



PCT\_US\_10\_579104\_ST25.txt  
SEQUENCE LISTING

<110> POLYPHOR LTD.  
Universität Zürich

<120> Template fixed beta-hairpin mimetics and their use in phage display

<130> P1338US

<140> PCT/US 10/579104  
<141> 2006-05-12

<150> PCT/EP 03/12783  
<151> 2003-11-15

<160> 44

<170> PatentIn version 3.5

<210> 1  
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<220>  
<223> Key sequence known to occur in Platelet-Derived Growth Factor (PDGF), see Ross, R.; Raines, E. W.; Bowden-Pope, D.F.; Cell, 1986, 46, 155-159.

<400> 1  
Val Arg Lys Lys  
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<210> 2  
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<223> Key sequence known to occur in Vasointestinal Peptide (VIP) showing neuroprotective properties against beta-amyloid neurotoxicity, see Proc. Natl. Am. Soc. USA, 1996, 96, 4143-4148.

<400> 2  
Lys Lys Tyr Leu  
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<210> 3  
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<220>  
<223> Key sequence known to occur in integrin alpha.sub4 beta.sub1, see Europ. J. Biol., 1996, 242, 352-362 and Int. J. Pept. Prot. Res., 1996, 47, 427-436.

<400> 3

Trp Leu Asp Val  
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<210> 4  
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<213> Artificial Sequence

<220>  
<223> Key sequence known to occur in Factor Xa inhibitors, see Al  
Obeidis, F.; Ostrem, J. A.; Drug Discovery Today, 1998, 3,  
223-231.

<400> 4

Tyr Ile Arg Leu Pro  
1 5

<210> 5  
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<220>  
<223> Key sequence known to occur in laminine, see EMBO. J., 1984, 3,  
1463.

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Tyr Ile Gly Ser Arg  
1 5

<210> 6  
<211> 5  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Key sequence known to occur in important physiologically active  
peptides, see Cell, 1987, 88, 989.

<400> 6

Ile Lys Val Ala Val  
1 5

<210> 7  
<211> 6  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Key sequence known to occur in important physiologically active  
peptides, see J. Biol. Chem., 1998, 273, 11001-11006 and  
11007-11011.

<220>

<221> misc\_feature  
 <222> (4)..(5)  
 <223> Xaa can be any naturally occurring amino acid  
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Pro Pro Arg Xaa Xaa Trp  
 1 5

<210> 8  
 <211> 10  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Hairpin mimetic derived from the general formula Cys-Z-Cys wherein the alpha amino group of the first amino acid is acetylated and wherein Z consists of 8 amino acids.

<220>  
 <221> DISULFID  
 <222> (1)..(10)

<220>  
 <221> MOD\_RES  
 <222> (1)..(1)  
 <223> ACETYLATION

<400> 8

Cys Lys Trp Phe Leu Ala His Tyr Ala Cys  
 1 5 10

<210> 9  
 <211> 14  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2 wherein the alpha amino group of the first amino acid is acetylated, wherein Z consists of 8 amino acids, and wherein both R1 and R2 consist of 2 amino acids.

<220>  
 <221> MOD\_RES  
 <222> (1)..(1)  
 <223> ACETYLATION

<220>  
 <221> DISULFID  
 <222> (3)..(12)

<400> 9

Glu Thr Cys Lys Trp Phe Leu Ala His Tyr Ala Cys Thr Lys  
 1 5 10

<210> 10  
 <211> 12  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> hairpin mimetic derived from the general formula Cys-Z-Cys wherein the alpha amino group of the first amino acid is acetylated and wherein Z consists of 10 amino acids.

<220>  
 <221> DISULFID  
 <222> (1)..(12)

<220>  
 <221> MOD\_RES  
 <222> (1)..(1)  
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<400> 10

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 1 5 10

<210> 11  
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 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2 wherein the alpha amino group of the first amino acid is acetylated, wherein Z consists of 10 amino acids, and wherein both R1 and R2 consist of 2 amino acids.

<220>  
 <221> MOD\_RES  
 <222> (1)..(1)  
 <223> ACETYLATION

<220>  
 <221> DISULFID  
 <222> (3)..(14)

<400> 11

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 1 5 10 15

<210> 12  
 <211> 12  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Hairpin mimetic derived from the general formula Cys-Z-Cys wherein the alpha amino group of the first amino acid is acetylated and wherein Z consists of 10 amino acids.

<220>  
 <221> DISULFID  
 <222> (1)..(12)

<220>  
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 <222> (1)..(1)  
 <223> ACETYLATION

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Cys Thr Lys Trp Phe Leu Ala His Tyr Ala Thr Cys  
 1 5 10

<210> 13  
 <211> 16  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2 wherein the alpha amino group of the first amino acid is acetylated, wherein Z consists of 10 amino acids, and wherein both R1 and R2 consist of 2 amino acids.

<220>  
 <221> MOD\_RES  
 <222> (1)..(1)  
 <223> ACETYLATION

<220>  
 <221> DISULFID  
 <222> (3)..(14)

<400> 13

Leu Glu Cys Thr Lys Trp Phe Leu Ala His Tyr Ala Thr Cys Lys Val  
 1 5 10 15

<210> 14  
 <211> 16  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2 wherein the alpha amino group of the first amino acid is acetylated, wherein Z consists of 10 amino acids, and wherein both R1 and R2 consist of 2 amino acids.

<220>  
 <221> MOD\_RES  
 <222> (1)..(1)  
 <223> ACETYLATION

<220>  
 <221> DISULFID  
 <222> (3)..(14)

&lt;400&gt; 14

Asn Gly Cys Thr Lys Trp Phe Leu Ala His Tyr Ala Thr Cys Lys Val  
 1 5 10 15

&lt;210&gt; 15

&lt;211&gt; 16

&lt;212&gt; PRT

&lt;213&gt; Artificial Sequence

&lt;220&gt;

<223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2  
 wherein the alpha amino group of the first amino acid is  
 acetylated, wherein Z consists of 10 amino acids, and wherein  
 both R1 and R2 consist of 2 amino acids.

&lt;220&gt;

&lt;221&gt; MOD\_RES

&lt;222&gt; (1)..(1)

&lt;223&gt; ACETYLATION

&lt;220&gt;

&lt;221&gt; DISULFID

&lt;222&gt; (3)..(14)

&lt;400&gt; 15

Gly Gly Cys Thr Lys Trp Phe Leu Ala His Tyr Ala Thr Cys Gly Gly  
 1 5 10 15

&lt;210&gt; 16

&lt;211&gt; 16

&lt;212&gt; PRT

&lt;213&gt; Artificial Sequence

&lt;220&gt;

<223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2  
 wherein the alpha amino group of the first amino acid is  
 acetylated, wherein Z consists of 10 amino acids, and wherein  
 both R1 and R2 consist of 2 amino acids.

&lt;220&gt;

&lt;221&gt; MOD\_RES

&lt;222&gt; (1)..(1)

&lt;223&gt; ACETYLATION

&lt;220&gt;

&lt;221&gt; DISULFID

&lt;222&gt; (3)..(14)

&lt;400&gt; 16

Glu Thr Cys Thr Lys Trp Phe Leu Ala His Tyr Ala Thr Cys Thr Lys  
 1 5 10 15

&lt;210&gt; 17

&lt;211&gt; 18

<212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2 wherein the alpha amino group of the first amino acid is acetylated, wherein Z consists of 10 amino acids, and wherein both R1 and R2 consist of 3 amino acids.

<220>  
 <221> MOD\_RES  
 <222> (1)..(1)  
 <223> ACETYLATION

<220>  
 <221> DISULFID  
 <222> (4)..(15)

<400> 17

Glu Leu Lys Cys Thr Lys Trp Phe Ser Asn His Tyr Gln Thr Cys Glu  
 1 5 10 15

Val Lys

<210> 18  
 <211> 18  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2 wherein the alpha amino group of the first amino acid is acetylated, wherein Z consists of 10 amino acids, and wherein both R1 and R2 consist of 3 amino acids.

<220>  
 <221> MOD\_RES  
 <222> (1)..(1)  
 <223> ACETYLATION

<220>  
 <221> DISULFID  
 <222> (4)..(15)

<400> 18

Lys Val Gly Cys Thr Lys Trp Phe Leu Ala His Tyr Ala Thr Cys Gly  
 1 5 10 15

Leu Glu

<210> 19  
 <211> 18  
 <212> PRT

<213> Artificial Sequence

<220>

<223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2 wherein the alpha amino group of the first amino acid is acetylated, wherein Z consists of 10 amino acids, and wherein both R1 and R2 consist of 3 amino acids.

<220>

<221> MOD\_RES

<222> (1)..(1)

<223> ACETYLATION

<220>

<221> DISULFID

<222> (4)..(15)

<400> 19

Gly Gly Gly Cys Thr Lys Trp Phe Leu Ala His Tyr Ala Thr Cys Gly  
1 5 10 15

Gly Gly

<210> 20

<211> 14

<212> PRT

<213> Artificial Sequence

<220>

<223> Hairpin mimetic derived from the general formula Cys-Z-Cys wherein the alpha amino group of the first amino acid is acetylated and wherein Z consists of 12 amino acids.

<220>

<221> DISULFID

<222> (1)..(14)

<220>

<221> MOD\_RES

<222> (1)..(1)

<223> ACETYLATION

<400> 20

Cys Gly Thr Lys Trp Phe Ser Asn His Tyr Gln Thr Gly Cys  
1 5 10

<210> 21

<211> 18

<212> PRT

<213> Artificial Sequence

<220>

<223> Hairpin mimetic derived from the general formula R1-Cys-Z-Cys-R2 wherein the alpha amino group of the first amino acid is acetylated, wherein Z consists of 12 amino acids, and wherein



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both R1 and R2 consist of 2 amino acids.

<220>  
<221> MOD\_RES  
<222> (1)..(1)  
<223> ACETYLATION

<220>  
<221> DISULFID  
<222> (3)..(16)

<400> 21

Glu Thr Cys Gly Thr Lys Trp Phe Ser Asn His Tyr Gln Thr Gly Cys  
1 5 10 15

Thr Lys

<210> 22  
<211> 8  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Core peptide sequence Z taken from the CDR L3 loop of an antibody described in Jiang, L. et al., Chimia, 2000,54, 558-563.

<400> 22

Leu Trp Tyr Ser Asn His Trp Val  
1 5

<210> 23  
<211> 8  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Modified core peptide sequence Z derived from core peptide sequence with the SEQ ID NO:22 containing a stabilizing beta-turn and a beta-sheet sequence according to Chou, P. Y., Fasman, G. D., J. Mol. Biol, 1977, 115, 135-175.

<400> 23

Lys Trp Phe Ser Asn His Tyr Gln  
1 5

<210> 24  
<211> 6  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Core peptide sequence Z constructed from peptide with the SEQ ID NO:25.

&lt;400&gt; 24

Phe Leu Ala His Tyr Ala  
 1 5

&lt;210&gt; 25

&lt;211&gt; 10

&lt;212&gt; PRT

&lt;213&gt; Artificial Sequence

&lt;220&gt;

<223> Oligopeptide which does not contain a dedicated stabilizing  
 beta-turn sequence or a beta-sheet sequence according to Chou, P.  
 Y., Fasman, G. D., J. Mol. Biol, 1977, 115, 135-175.

&lt;400&gt; 25

Leu Trp Tyr Ser Asn His Trp Val Lys Trp  
 1 5 10

&lt;210&gt; 26

&lt;211&gt; 39

&lt;212&gt; DNA

&lt;213&gt; Artificial Sequence

&lt;220&gt;

<223> Oligonucleotide No. 1 used to construct insert DNA coding for  
 template fixed hairpin mimetic of SEQ ID NO:10 and used to  
 construct insert DNA coding for randomized library template fixed  
 beta-hairpin mimetics having sequences according to SEQ ID NO:42.

&lt;400&gt; 26

catgcccggg tacctttcta ttctcactct gaaacctgc

39

&lt;210&gt; 27

&lt;211&gt; 84

&lt;212&gt; DNA

&lt;213&gt; Artificial Sequence

&lt;220&gt;

<223> Oligonucleotide No. 2 used to construct insert DNA coding for  
 template fixed hairpin mimetic of SEQ ID NO:10.

&lt;400&gt; 27

catgtttcgg ccgagccacc acctttggtg caggtctgat aatggttgct gaaccatttg

60

gtgcaggttt cagagtgaga atag

84

&lt;210&gt; 28

&lt;211&gt; 30

&lt;212&gt; DNA

&lt;213&gt; Artificial Sequence

&lt;220&gt;

<223> DNA sequence coding for the peptide shown in SEQ ID NO:8.

&lt;400&gt; 28

tgcaaattgg ttctggcgca ttatgcgtgc

30

<210> 29  
 <211> 42  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:9.  
  
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 gaaacctgca aatgggttcct ggcgcattat gcgtgcacca aa 42

<210> 30  
 <211> 36  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:10.  
  
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 tgcaccaaat ggttcagcaa ccattatcag acctgc 36

<210> 31  
 <211> 48  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:11.  
  
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 gaaacctgca ccaaatgggtt cagcaaccat tatcagacct gcaccaa 48

<210> 32  
 <211> 36  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:12.  
  
 <400> 32  
 tgcaccaaat ggttcctggc gcattatgcg acctgc 36

<210> 33  
 <211> 48  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:13.  
  
 <400> 33  
 ctggaatgca ccaaatgggtt cctggcgcatt tatgcgacct gcaaagtt 48

<210> 34  
 <211> 48

<212> DNA  
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 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:14.  
  
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 aacggttgca ccaaatggtt cctggcgcac tatgcgacct gcaaagtt 48  
  
 <210> 35  
 <211> 48  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:15.  
  
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 ggtggttgca ccaaatggtt cctggcgcac tatgcgacct gcggcggt 48  
  
 <210> 36  
 <211> 48  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:16.  
  
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 gaaacctgca ccaaatggtt cctggcgcac tatgcgacct gcacccaaa 48  
  
 <210> 37  
 <211> 54  
 <212> DNA  
 <213> Artificial Sequence  
  
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 <223> DNA sequence coding for the peptide shown in SEQ ID NO:17.  
  
 <400> 37  
 gaactgaaat gcaccaaatt gttcagcaac cattatcaga cctgcgaagt taaa 54  
  
 <210> 38  
 <211> 54  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> DNA sequence coding for the peptide shown in SEQ ID NO:18.  
  
 <400> 38  
 aaagttggtt gcaccaaatt gttcctggcg cattatgcga cctgcggtct ggaa 54  
  
 <210> 39  
 <211> 54  
 <212> DNA  
 <213> Artificial Sequence

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<220>
<223> DNA sequence coding for the peptide shown in SEQ ID NO:19.

<400> 39
gggtggtggct gcaccaaata gttcctggcg cattatgcga cctgcggcgg tggt          54

<210> 40
<211> 42
<212> DNA
<213> Artificial sequence

<220>
<223> DNA sequence coding for the peptide shown in SEQ ID NO:20.

<400> 40
tgcggtacca aatggttcag caaccattat cagaccggtt gc          42

<210> 41
<211> 54
<212> DNA
<213> Artificial sequence

<220>
<223> DNA sequence coding for the peptide shown in SEQ ID NO:21.

<400> 41
gaaacctgcg gtaccaaata gttcagcaac cattatcaga ccggttgcac caaa          54

<210> 42
<211> 48
<212> DNA
<213> Artificial sequence

<220>
<223> DNA sequence of randomized template fixed beta-hairpin mimetic
      Phage library.

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<223> n is a, c, g, or t

<220>
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<221> misc_feature
<222> (16)..(17)
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<220>
<221> misc_feature
<222> (28)..(29)
<223> n is a, c, g, or t

<220>
<221> misc_feature

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<222> (31)..(32)  
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 <223> n is a, c, g, or t

<220>  
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 <222> (37)..(38)  
 <223> n is a, c, g, or t

<400> 42  
 gaaacctgcn nknknknkcgcg tgggtgacnnk nnknknknkt gcaccaaa

48

<210> 43  
 <211> 16  
 <212> PRT  
 <213> Artificial Sequence

<220>  
 <223> Translated protein sequence of a randomized template fixed  
 beta-hairpin mimetic phage library

<220>  
 <221> DISULFID  
 <222> (3)..(14)

<220>  
 <221> MISC\_FEATURE  
 <222> (4)..(6)  
 <223> Xaa can be any naturally occurring amino acid

<220>  
 <221> MISC\_FEATURE  
 <222> (10)..(13)  
 <223> Xaa can be any naturally occurring amino acid

<400> 43

Glu Thr Cys Xaa Xaa Xaa Arg Gly Asp Xaa Xaa Xaa Xaa Cys Thr Lys  
 1 5 10 15

<210> 44  
 <211> 84  
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 <213> Artificial Sequence

<220>  
 <223> Oligonucleotide No. 3 used to construct insert DNA coding for  
 randomized library template fixed beta-hairpin mimetics having  
 sequences according to SEQ ID NO:42.

<220>  
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 <223> n is a, c, g, or t

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<220>  
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 <223> n is a, c, g, or t

<220>  
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 <222> (61)..(62)  
 <223> n is a, c, g, or t

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 nngcaggttt cagagtgaga atag 84